

ATPase ACTIVITY AND ITS CONNECTION WITH THE ACTIVE K^+ UPTAKE OF DIFFERENT SEGMENTS OF RICE AND WHEAT ROOTS

ILDIKÓ TÓTH and F. ZSOLDOS

Department of Plant Physiology, Attila József University, Szeged
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Summary

Microsomal ATPase activity of different segments of a thermophilic plant rice (*Oryza sativa* L. cv. Dunghan Shali) and a non-thermophilic plant wheat (*Triticum aestivum* L. cv. Rannaja-12) were investigated. Seedlings were grown in water culture (5×10^{-4} M $CaSO_4$ solution) under controlled conditions. ATPase activities of both plants were similar and could be activated by adding 5×10^{-4} M KCl and 5×10^{-4} M $CaCl_2$, but there were differences between the ATPase pattern of the species along the primary roots. In the case of rice, there were large enzyme activities in the elongation zone, while regarding the wheat a determined peak could be measured at the third segment from the root tip. ATPase activities of the first and the last portions (4—5 cm) as well as those of the second and the fourth zones were similar. Our results agree with earlier data on the K^+ uptake and of the K^+ content experiments.

Introduction

In our preliminary studies some enzymkinetic parameters of rice root (e. g. optimum protein content, ATP concentration, ion strenght, pH) were determined (TÓTH, 1976). Later similar measurements were done using ATPase enzyme of wheat roots, too. Our results which correspond to the literature data prove the important role of ATPase enzyme in active ion uptake processes of plants (CHRISTIANSEN and LINDBERG, 1976; LEIGHT and WYN JONES, 1976; KUIPER et al, 1974). There is a strong correlative evidence relating to K^+ uptake to K^+ stimulating effect of the microsomal ATPase of oat root (BALKE and HODGES, 1975). In the case of maize LEONARD and HOTCHKISS (1976) reported similar result, and using wheat root KYLIN and KHÄR (1976) demonstrated this evidence.

Recently, the connection between the active cation uptake and cation activated ATPases can be considered as a proof. It is remarkable that almost in all cases these investigations had been done with roots of different length but intact. Earlier studies, however, indicated non-equal participation of the different zones of the primary roots of rice and wheat in the K^+ uptake and significant differences could be measured in the K^+ content of the individual segments, too. (ZSOLDOS and KARVALY, 1978). Furthermore, there are variances among different species in respect of ion uptake and the anomalous high K^+ uptake of thermophilic plants at low temperature may be explained by this.

In the present work the main aim was to examine the microsomal ATPase activity and activation of each segment along the primary roots. The last question is very interesting because in our earlier studies the (Ca^{2+} , K^{+}) ATPase activity seemed to be under genetic control and appears only on a determined level of the organisation (ERDEI et al, 1977). The first, 4–5 mm long segment of the primary root (division zone) corresponds to the lower level of organisation, while the more distant portions from the root tip (elongation, absorption zones) can be considered to be of higher organisational status.

Our present work is considered to be a preliminary study of a series of investigations in which transport features, hormone sensitivity of different segments of roots will be investigated.

Materials and Methods

In our experiments with ATPase activity of 1 cm portion of roots of rice and wheat were investigated in parallel. Seedlings were grown under standard conditions, in low salt ($5 \times 10^{-4} \text{M}$ CaSO_4) solution, determined light, and humidity, as described earlier (ERDEI et al, 1977). The plants used in the experiments were 5–7 days old, their roots being about 6–7 cm long. The roots were washed three times in distilled water then cut to 1 cm long segments. Preparation of microsomal fraction (10,000–30,000 g) was as described earlier, too, using an extraction medium containing 250 mM sucrose, 25 mM Tris-HCl buffer and 3 mM EDTA, pH value was 6.9 in the case of rice and 7.5 using wheat plant, respectively.

The determination of ATPase activity was done using a reaction system having 1 ml total volume which contained 100–200 μg protein, 5 mM ATP, 5 mM KCl, 0.5 mM CaCl_2 and 20 mM Tris-HCl buffer (pH 6.9 and 7.5 respectively). The reaction was started by addition of ATP run for 10 minutes at 32 °C and stopped with ice cold 40% TCA. The specific activity of enzyme was calculated as $\mu\text{mole P}_i$ released by 1 mg protein per hour. The classical FISKE and SUBBAROW method (1925) was used for the determination of P_i and the protein was assayed according to the LOWRY method (1951). All experiments were made on triplicate, sometimes five times, and the error was about 5%.

Results and Discussion

Figures 1. and 2. show the ATPase activity of different segments of rice and wheat roots. From the data it is visible that the activities are in the same order. KYLIN and KHÄR (1974) reported similar results using wheat roots. To compare the two Figures with each other the relative low enzyme activity of the first and the last segments can be established. This almost equal correspondence is very interesting since the first segment contains mostly rapidly dividing cells having minimum vacuola, while in the last zone the vessels have already appeared and a determined state of root hair can be detected. In the case of rice the ATPase activity between the 2–5 segments is almost similar and it is interesting enough that it is 80% more than the activities of the first and the last portions. As regards the wheat, a significant peak can be detected in the third zone which activity is also 80% more than in the first, while 68% higher than at the second and the fourth segments.

Differences in the ATPase pattern of the two species may be ascribed to differences which are in the morphological, tissue and cell levels of roots of the thermophilic and non-thermophilic plants (ZSOLDOS and GULYÁS, 1979). Data measured at the wheat completely equal the K^{+} uptake pattern of different segment at 25 °C, since the K^{+} content at the third segments is a manifold of the amount of the first and the last portions (ZSOLDOS and KARVALY, 1978). In the case of rice a gradual rise

can be seen in the K^+ uptake, there are higher amounts at the more mature segments of root. The first segments of both plants have the lowest K^+ uptake.

Comparing the results to data of content of different segments another interesting connection can be observed (ZSOLDOS and KARVALY, 1978). The more differentiated segments of both plants show decreasing levels, the first zones having significantly more K^+ content. As regards the (Ca^{2+}, K^+) ATPase patterns, the K^+ uptake curves and the K^+ content distributions there may be a strong evidence that the uptake is connected with the function of transport enzyme. This connected process is most determined in the elongation zone, while the first segment has only a small role in the ion uptake process of root.

From the data above it seems that the different segments of various plants show important unequal features in the transport processes. Our earlier studies indicated that the K^+ uptake and the (Ca^{2+}, K^+) ATPase activity is under hormonal control (ERDEI et al, 1979). So it may be a very interesting question to investigate the hormone and the herbicide sensitivity of different segments of roots.

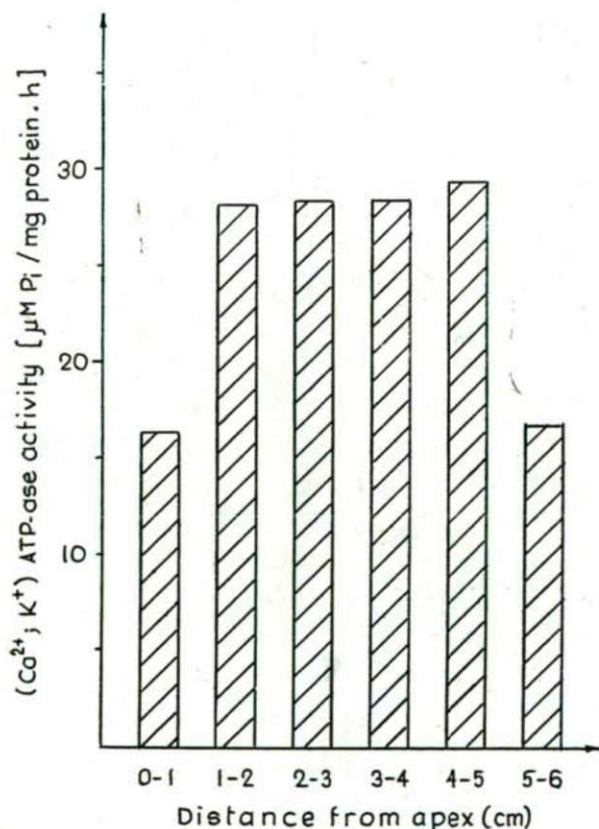


Fig. 1. Microsomal ATPase activity of different segments of rice along the primary roots.

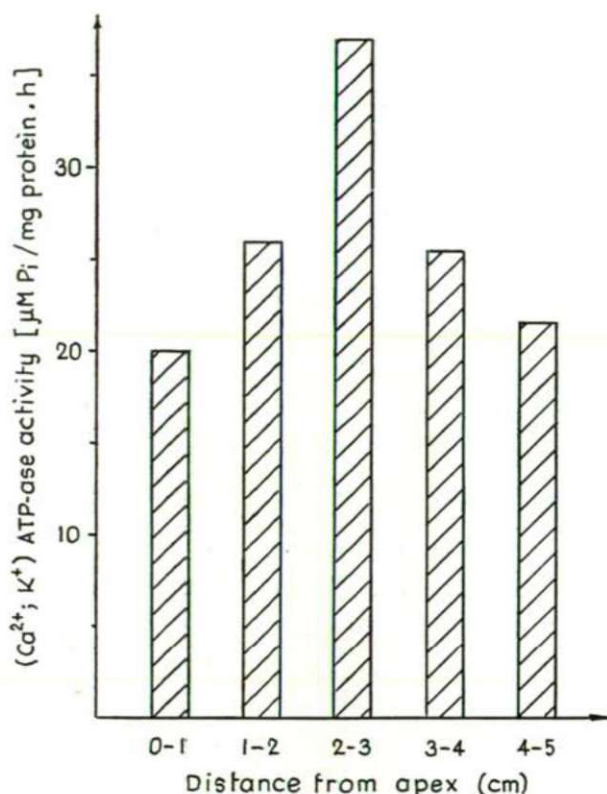


Fig. 2. Microsomal ATPase activity of different segments of wheat along the primary roots.

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References

- BALKE, N. E.—HODGES, T. K. (1975): Plasma membrane adenosine triphosphatase of oat roots. — *Plant Physiol.* 55, 83—86.
- CHRISTIANSEN, J. L.—LINDBERG, S. (1976): Kinetic studies of (Mg²⁺)ATP-ase in oat roots. — *Physiol. Plant.* 36, 110—112.
- ERDEI, L.—ZSOLDOS, F. (1977): Potassium absorption by rice at different levels of organisation. I. Effects of temperature and calcium on K⁺ fluxes and content. — *Ibid* 41, 99—104.
- ERDEI, L.—TÓTH, I.—ZSOLDOS, F. (1977): II. ATP-ase activity in roots, calluses and redifferentiated roots. — *Ibid* 41, 105—108.
- ERDEI, L.—TÓTH, I.—ZSOLDOS, F. (1979): Hormonal regulation of Ca²⁺-stimulated K⁺ influx and Ca²⁺, K⁺-ATP-ase in rice roots: in vivo and in vitro effects of auxins and reconstitution of the ATP-ase. — *Ibid* 45, 448—452.
- FISKE, C. H.—SUBBAROW, Y. (1925): The colorimetric determination of phosphorus. — *J. Biol. Chem.* 66, 375—400.

- HENDRIX, D. L.—KENNEDY, R. M. (1977): Adenosine triphosphatase from soybean callus and root cells. — *Plant Physiol.* 59, 264—267.
- KÄHR, M.—KYLIN, A. (1974): Effects of divalent cations and oligomycin on membrane ATP-ase from roots of wheat and oat in relation to salt status and cultivating. — *in* *Membrane transport in plants*. (U. Zimmermann — J. Dainty) — 321—325.
- KUIPER, P. J. C.—KÄHR, M.—STUIVER, C. E. E.—KYLIN, A. (1974): Lipid composition of whole roots and of Ca^{2+} Mg^{2+} -activated adenosine triphosphatases from wheat and oat as related to mineral nutrition. — *Physiol. Plant.* 32, 33—36.
- LEIGHT, R. A.—WYN JONES, R. G. (1975): Correlations between ion-stimulated adenosine triphosphatase activities and ion influx in maize roots. — *J. Exp. Bot.* 26, 508—520.
- LEONARD, R. T.—HODGES, T. K. (1973): Characterisation of plasma membrane-associated adenosine triphosphatase activity of oat roots. — *Plant Physiol.* 52, 6—12.
- LEONARD, R. T.—HOTCHKISS, C. W. (1976): Cation-stimulated adenosine triphosphatase activity and cation transport in corn roots. — *Ibid* 58, 331—335.
- LOWRY, O. H.—ROSEBROUGH, N. J. (1951): Protein measurement with the folin phenol reagent. *J. Biol. Chem.* 193, 265—275.
- TÓTH, I.—ZSOLDOS, F. (1976): Effects of Saturn-50 and Synpran-111 herbicides on the membrane-bound ATP-ase activity of roots and young shoots of rice. — *Acta Biol. Szeged*, 22, 73—78.
- TÓTH, I. (1976): Összehasonlító vizsgálatok rizsgyökér ATP-áz enzimmel. — doktori értekezés, p. 94.
- ZSOLDOS, F.—KARVALY, B. (1978): Effects of Ca^{2+} and temperature on potassium uptake along roots of wheat, rice and cucumber. — *Physiol. Plant.* 43, 326—330.
- ZSOLDOS, F.—KARVALY, B. (1978): Effects of Ca^{2+} and temperature on K^+ content distribution along roots of wheat, rice and cucumber. — *Ibid* 43, 331—336.
- ZSOLDOS, F.—GULYÁS, S. (1979): Changes induced by chilling in the ion uptake, growth and anatomical structure of rice roots. — *Acta Biol. Szeged*, 25, 69—76.

Address of the authors:

Dr. ILDIKÓ TÓTH

Dr. F. ZSOLDOS

Department of Plant Physiology
A. J. University, H-6701 Szeged,
P. O. Box 428. Hungary